CLAIMS

WHAT IS CLAIMED IS:

1. A fluorescent reporter compound of the formula:

Z-L-Cy

wherein

5

Z is a nucleotide;

L is a linker of sufficient length to connect the nucleotide derivative to the cyanine dye, such that the cyanine dye does not significantly interfere with the overall binding and recognition of the nucleotide derivative by a nucleic acid replication enzyme; and

Cy is a cyanine dye of the formula:

 R_4 A +N R_1 $(CH_2)_n$ R_3 R_2

15

wherein

A and B are each independently the atoms necessary to form a cyanine nuclei;

R₁ and R₂ are each independently C₁-C₆ alkyl;

R₃ is hydrogen, or C₁-C₄ alkyl;

20

 R_4 and R_5 are each independently selected from the group consisting of H and SO_3 ; and

n is 2, 3, or 4.

2. A compound according to Claim 1 wherein R_4 and R_5 are both SO_3 .

PATENT

- 3. A compound according to Claim 1 wherein R_1 and R_2 are both $C_1\text{-}C_4$ alkyl.
- 4. A compound according to Claim 1 wherein R_1 and R_2 are identical.
- 5. A compound according to Claim 1 of the formula:

wherein

5

X is O, S, NR9, or CR9R10;

R9 and R10 are each independently H or C1-C4 alkyl; and

Y is a diradical moiety having 3 to 20 atoms, at least three of which include an alkynyl group and one or more heteroatoms.

6. A compound according to Claim 1 of the formula:

$$Z$$
 NH
 $(CH_2)_m$
 R_4
 C
 N_{\uparrow}
 R_1
 R_3

15 wherein:

C and D represent ring structures with sufficient carbon atoms to make up a benzene or naphthalene ring; and

m is an integer from 1 to 6.

- 7. A compound according to Claim 6 wherein R4 and R5 are both SO3.
- 5 8. A compound according to Claim 6 wherein R₁ and R₂ are both C₁-C₄ alkyl.
 - 9. A compound according to Claim 8 wherein R₁ and R₂ are identical.
 - 10. A compound according to Claim 1 of the formula:

10

11. A compound according to Claim 1 of the formula:

15 12. A compound according to Claim 1 having a fluorescence maxima greater than 750 nm as

measured in an aqueous solution.

13. A fluorescent reporter compound of the formula:

5 wherein

Z is a nucleotide derivative;

L is a linker having a chain length of at least 8 atoms; and

Cy is a cyanine dye of the formula:

$$R_4$$
 A
 $+N$
 R_1
 $(CH_2)_n$
 R_3
 R_2

10

15

wherein

A and B are each independently the atoms necessary to form a cyanine nuclei;

 R_1 and R_2 are each independently $C_1\text{-}C_6$ alkyl;

R₃ is hydrogen, or C₁-C₄ alkyl;

 R_{4} and R_{5} are each independently selected from the group consisting of H and SO_{3} ; and

n is 2, 3, or 4.

14. A compound according to Claim 13 of the formula:

$$Z$$
 NH
 C
 NH
 $(CH_2)_m$
 R_4
 C
 N_1
 R_2
 R_3

wherein:

C and D represent ring structures with sufficient carbon atoms to make up a benzene or naphthalene ring; and

m is an integer between and including 1-6.

- 15. A compound according to Claim 13 having a fluorescence maxima greater than 750 nm as measured in an aqueous solution.
- 16. A method of nucleic acid sequence analysis comprising:
- reacting a fluorescent reporter labeled compound according to Claim 1 with a first nucleic acid sequence to produce a second nucleic acid sequence labeled with the fluorescent reporter-labeled compound; and

detecting the reporter on the second nucleic acid sequence.

- 17. A method for determining the base sequence of DNA comprising:
- providing a mixture of fluorescent reporter labeled compounds according to Claim 1 corresponding to each of the four DNA bases;

reacting a DNA template with a replication enzyme, a mixture of DNA nucleotides, and the mixture of fluorescent reporter-labeled compounds;

producing DNA fragments having a fluorescent reporter-labeled compound covalently attached to the 3'-terminal residue of each DNA fragment;

separating the fluorescent reporter-labeled DNA fragments; and

20

PATENT

detecting the reporter for each separated fluorescent reporter-labeled DNA fragment thereby identifying the DNA sequence.

18. A compound of the formula:

$$\begin{array}{c|c} & \text{NCS} \\ (CH_2)_p \\ \hline \\ R_1 \\ (CH_2)_n \\ R_3 \\ R_2 \\ \end{array}$$

wherein

5

10

15

A and B are each independently the atoms necessary to form a cyanine nuclei;

X is O, S, NR₉, or CR₉R₁₀;

R₁ and R₂ are each independently C₁-C₆ alkyl;

R₃ is hydrogen, or C₁-C₄ alkyl;

R₄ and R₅ are each independently selected from the group consisting of H and SO₃, provided that at least one of R₄ and R₅ is SO₃;

R9 and R10 are each independently H or C1-C4 alkyl;

p is an integer between and including 2-8; and

n is an integer between and including 2-4.

- 19. A compound according to Claim 18 wherein R₄ and R₅ are both SO₃.
- 20. A compound according to Claim 18 wherein R1 and R2 are both C1-C4 alkyl.
- 21. A compound according to Claim 18 wherein R_1 and R_2 are identical.
- 20 22. A compound according to Claim 18 of the formula:

22. A compound according to Claim 18 of the formula:

$$\begin{array}{c} \text{NCS} \\ \text{(CH}_2)p \\ \\ \text{N} \\ \\ \text{R}_1 \end{array}$$

5